7

# **WEST Search History**

Hide Items Restore Clear Cancel

DATE: Friday, July 09, 2004

Hide?	<u>Set</u> Name	Query	<u>Hit</u> <u>Count</u>					
DB=PGPB, USPT, USOC, EPAB, JPAB, DWPI; PLUR=YES; OP=ADJ								
	L1	oligo-dT near\$5 primer	0					
<b></b>	L2	oligo-dT	3593					
	L3	L2 near (primer or sequence)	1369					
	L4	L3 near (detectable marker or label or fluorescen\$ or fluorophor\$)	2					
	L5	L3 same (detectable marker or label or fluorescen\$ or fluorophor\$)	75					
	L6	L2 same (detectable marker or label or fluorescen\$ or fluorophor\$)	133					
	L7	L2 near (tag or marker)	1					
	L8	L2 same (tag or marker)	147					
	L9	L3 same (tag or marker)	69					
	L10	reverse Transcrib\$	10356					
	L11.	L10 and (16 or 17)	72					
	L12	L10 and (15 or 18 or 19)	111					
	L13	(111 or 112) and (restriction endonuclease or endonuclease or restriction enzyme or sequence-specific cleav\$ agent)	100					
	L14	(ligat\$ same adapter same promoter)	937					
Г	L15	113 and 114	8					
	L16	L15 and amplif\$	8					
	L17	(selective near PCR) or (selective near polymerase chain reaction) or (selective amplification)	3153					
	L18	(selective near linear amplification)	2					
1,000	L19	linear amplification	3095					
	L20	linear PCR	127					
	L21	differential display	2802					
	L22	L16 and 117	Ó					
	L23	113 and (adapter same promoter)	9					
	L24	113 and (adapter and promoter)	44					
	L25	(123 or 124) and (117 or 119)	18					
	L26	L25 and l21	0					
	L27	L24 and l21	3					
	L28	L13 and l21	22					
	L29	L28 and (adaptor or adapter)	5					

## END OF SEARCH HISTORY

\* \* \* \* \* \* \* \* Welcome to STN International NEWS 1 Web Page URLs for STN Seminar Schedule - N. America NEWS "Ask CAS" for self-help around the clock NEWS 3 May 12 EXTEND option available in structure searching NEWS 4 Polymer links for the POLYLINK command completed in REGISTRY May 12 NEWS 5 May 27 New UPM (Update Code Maximum) field for more efficient patent SDIs in CAplus NEWS May 27 CAplus super roles and document types searchable in REGISTRY NEWS Jun 22 STN Patent Forums to be held July 19-22, 2004 NEWS Jun 28 Additional enzyme-catalyzed reactions added to CASREACT NEWS 9 Jun 28 ANTE, AQUALINE, BIOENG, CIVILENG, ENVIROENG, MECHENG, and WATER from CSA now available on STN(R) NEWS EXPRESS MARCH 31 CURRENT WINDOWS VERSION IS V7.00A, CURRENT MACINTOSH VERSION IS V6.0c(ENG) AND V6.0Jc(JP), AND CURRENT DISCOVER FILE IS DATED 26 APRIL 2004 NEWS HOURS STN Operating Hours Plus Help Desk Availability NEWS INTER General Internet Information NEWS LOGIN Welcome Banner and News Items Direct Dial and Telecommunication Network Access to STN NEWS PHONE NEWS WWW CAS World Wide Web Site (general information) Enter NEWS followed by the item number or name to see news on that specific topic. All use of STN is subject to the provisions of the STN Customer agreement. Please note that this agreement limits use to scientific research. Use for software development or design or implementation of commercial gateways or other similar uses is prohibited and may result in loss of user privileges and other penalties. FILE 'HOME' ENTERED AT 13:15:31 ON 09 JUL 2004 => file .biotech caplus COST IN U.S. DOLLARS SINCE FILE TOTAL ENTRY SESSION FULL ESTIMATED COST 0.21 0.21 FILES 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS' ENTERED AT 13:16:11 ON 09 JUL 2004 ALL COPYRIGHTS AND RESTRICTIONS APPLY. SEE HELP USAGETERMS FOR DETAILS. 7 FILES IN THE FILE LIST => s kane M?/au or hagel A?/au or dombkowski A?/au 2912 KANE M?/AU OR HAGEL A?/AU OR DOMBKOWSKI A?/AU => s l1 and differential display 0 L1 AND DIFFERENTIAL DISPLAY => s l1 and express##### gene 0 L1 AND EXPRESS##### GENE => s l1 and mRNA 56 L1 AND MRNA => s l4 and diffential?

0 L4 AND DIFFENTIAL?

```
=> s l1 and express? mRNA
             0 L1 AND EXPRESS? MRNA
=> s differential display or (differentially expressed (5a) mRNA)
         20308 DIFFERENTIAL DISPLAY OR (DIFFERENTIALLY EXPRESSED (5A) MRNA)
=> s 11 and 17
1.8
             0 L1 AND L7
=> d 17 and (oligo-dT (5a) (primer or sequenc?))
'(OLIGO-DT, (5A) ' IS NOT VALID HERE
=> s 17 and (oligo-dT (5a) (primer or sequenc?))
           128 L7 AND (OLIGO-DT (5A) (PRIMER OR SEQUENC?))
=> s 19 and (detectable marker or marker or tag or label or fluorescen or
fluorophor)
) IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter
"HELP COMMANDS" at an arrow prompt (=>).
=> s 19 and reverse transcrib
=>
=>
=>
<---->
=> s 19 and (reverse transcrib)
) IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter
"HELP COMMANDS" at an arrow prompt (=>).
=> d his
     (FILE 'HOME' ENTERED AT 13:15:31 ON 09 JUL 2004)
     FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS'
     ENTERED AT 13:16:11 ON 09 JUL 2004
L1
           2912 S KANE M?/AU OR HAGEL A?/AU OR DOMBKOWSKI A?/AU
L2
              0 S L1 AND DIFFERENTIAL DISPLAY
L3
              O S L1 AND EXPRESS##### GENE
L4
             56 S L1 AND MRNA
L5
              0 S L4 AND DIFFENTIAL?
L6
              0 S L1 AND EXPRESS? MRNA
          20308 S DIFFERENTIAL DISPLAY OR (DIFFERENTIALLY EXPRESSED (5A) MRNA)
L7
1.8
              0 S L1 AND L7
            128 S L7 AND (OLIGO-DT (5A) (PRIMER OR SEQUENC?))
=> s 19 and (reverse trascrib?)
L10
            0 L9 AND (REVERSE TRASCRIB?)
=> s 19 and adaptor
             1 L9 AND ADAPTOR
=> d d his
D NOT VALID FOR HISTORY RANGE
You may enter one of the following:
  1. L1- to display your entire session history
  2. A specific L-number or range of L-numbers to be displayed
  3. L# to display the most recently created L-number
  4. A number, N, to display the N most recent L-numbers
```

ENTER (L1-), L#, OR ?:end

=> 111

L11 IS NOT A RECOGNIZED COMMAND

The previous command name entered was not recognized by the system. For a list of commands available to you in the current file, enter "HELP COMMANDS" at an arrow prompt (=>).

=> d his 111 all

'ALL' IS NOT VALID HERE

For an explanation, enter "HELP DISPLAY HISTORY".

=> d l11 all

L11 ANSWER 1 OF 1 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN

AN 95:703492 SCISEARCH

GA The Genuine Article (R) Number: RY760

TI DESCRIPTION OF THE ENTIRE MESSENGER-RNA POPULATION BY A 3'-END CDNA FRAGMENT GENERATED BY CLASS IIS RESTRICTION ENZYMES

AU KATO K (Reprint)

CS JRDC, ERATO, OKAYAMA CELL SWITCHING PROJECT, SAKYO KU, 103-5 TANAKAMONZENCHO, KYOTO 606, JAPAN (Reprint)

CYA JAPAN

SO NUCLEIC ACIDS RESEARCH, (25 SEP 1995) Vol. 23, No. 18, pp. 3685-3690. ISSN: 0305-1048.

DT Article; Journal

FS LIFE

LA ENGLISH

REC Reference Count: 17

AB A novel means of recording the expression status of the total gene population is described, Digestion of cDNA by class IIS restriction enzymes produces a fragment with a poly (A) stretch and a 5' overhang with an unknown sequence, This fragment contains information such as the class IIS enzyme that cuts cDNA nearest to the poly (A) stretch, the sequence of the 5' overhang, and the size of the fragment. Expressed genes can be discriminated and displayed by the fragment as follows: (i) cut the cDNA with one class IIS restriction enzyme; (ii) ligate the digested cDNA to that from a pool of 64 biotinylated adaptors cohesive to all possible overhangs; (iii) digest by other two class IIS enzymes; (iv) recover the ligated molecules with streptavidin-coated paramagnetic beads; (v) perform PCR with the adaptor-primer and an anchored oligo-dT primer; (vi) separate the amplified fragments by denaturing polyacrylamide gel electrophoresis. Repeat the experiment with 64 adaptors, three enzymes and three anchored oligo-dT

fragments by denaturing polyacrylamide gel electrophoresis. Repeat the experiment with 64 adaptors, three enzymes and three anchored oligo-dT primers displays most of the expressed genes, Because redundancy is minimized, this technique is also ideal for generating tags for expressed genes, with which to construct a transcript map of the genome.

CC BIOCHEMISTRY & MOLECULAR BIOLOGY

STP KeyWords Plus (R): DIFFERENTIAL DISPLAY; PCR; RNA

RF 93-0362 001; LANGUAGE IMPLEMENTATION; INTEGRATED MEDICAL WORKSTATION; HYPERTEXT SYSTEM; PROGRAMMING ENVIRONMENTS 93-1577 001; DNA RESTRICTION; SITE-SPECIFIC ENDONUCLEASE; COLLECTION OF ANNOTATED PROTEIN SEGMENTS; SBASE DOMAIN LIBRARY; ENZYME DATA-BANK 93-4847 001; HETEROLOGOUS EXPRESSION; CHROMOSOMAL DNA; GENE ENCODING

METHYLMALONYL-COENZYME-A MUTASE

KE				
Referenced Author	Year	VOL	PG	Referenced Work
(RAU)	(RPY)	(RVL)	(RPG)	(RWK)
=======================================	+=====.	+=====	+======	+====================================
ADAMS M D	1991	21	1651	SCIENCE
BRENNER S	1989	86	8902	P NATL ACAD SCI USA
DUNBAR B S	1987		İ	2 DIMENSIONAL ELECTR
GUBLER U	1983	25	263	GENE
HORTON R M	1994	16	42	BIOTECHNIQUES
KATO K	1990	2	704	EUR J NEUROSCI
		•	•	•

KATO K	1993	6	1	GENOME ANAL GENOME M
KERNIGHAN B W	1984	İ	j	UNIX PROGRAMMING ENV
KO M S H	1990	18	5705	NUCLEIC ACIDS RES
LIANG P	1993	21	3269	NUCLEIC ACIDS RES
LIANG P	1992	257	967	SCIENCE
SAMBROOK J	1989			MOL CLONING LABORATO
SMITH D R	1992	2	21	PCR METH APPL
SOUTHERN E M	1992	2	412	CURRENT OPINION GENE
SZYBALSKI W	1991	100	13	GENE
UNRAU P	1994	145	163	GENE
WELSH J	1992	20	4965	NUCLEIC ACIDS RES

=> d hi

'HI' IS NOT A VALID FORMAT FOR FILE 'SCISEARCH'

The following are valid formats:

The default display format is BIB.

ABS ---- AB

ALL ---- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC, AB, CC, ST, STP, RF, RE

BIB ---- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC

CBIB --- CBIB

IABS --- AB

IALL --- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC, AB, CC, ST, STP, RF, RE

IBIB --- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC

IND ---- CC, ST, STP, RF

SAM ---- AN, GA, TI, REC, CC, ST, STP, RF

SCAN --- AN, GA, TI, CC, ST, STP, RF (random display without answer numbers)

TRI ---- AN, GA, TI, REC, CC, ST, STP, RF

HIT ---- All fields containing hit terms

KWIC --- All hit terms plus 20 words on either side

OCC ---- List of display fields containing hit terms

Hit terms will be highlighted in all displayable fields except PY.

To display a particular field or fields, enter the display field codes. For a list of display field codes, enter 'HELP DFIELDS' at an arrow prompt (=>). Examples of formats include: 'BIB'; 'AB'; 'SO,ST'. You may specify the format fields in any order, and the information will be displayed in the same order as the format specification.

The same formats (except for HIT, KWIC, OCC, and SCAN) may be used with the DISPLAY ACC command to display the record for a specified Accession Number.

ENTER DISPLAY FORMAT (BIB):d his
'D' IS NOT A VALID FORMAT FOR FILE 'SCISEARCH'
'HIS' IS NOT A VALID FORMAT FOR FILE 'SCISEARCH'

The following are valid formats:

The default display format is BIB.

```
ABS ---- AB
```

ALL ---- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC, AB, CC, ST, STP, RF, RE

BIB ---- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC

CBIB --- CBIB

IABS --- AB

IALL --- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC, AB, CC, ST, STP, RF, RE

IBIB --- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC

IND ---- CC, ST, STP, RF

SAM ---- AN, GA, TI, REC, CC, ST, STP, RF

SCAN --- AN, GA, TI, CC, ST, STP, RF (random display without answer numbers)
TRI ---- AN, GA, TI, REC, CC, ST, STP, RF
HIT ---- All fields containing hit terms
KWIC --- All hit terms plus 20 words on either side
OCC ---- List of display fields containing hit terms

Hit terms will be highlighted in all displayable fields except PY.

To display a particular field or fields, enter the display field codes. For a list of display field codes, enter 'HELP DFIELDS' at an arrow prompt (=>). Examples of formats include: 'BIB'; 'AB'; 'SO,ST'. You may specify the format fields in any order, and the information will be displayed in the same order as the format specification.

The same formats (except for HIT, KWIC, OCC, and SCAN) may be used with the DISPLAY ACC command to display the record for a specified Accession Number.

ENTER DISPLAY FORMAT (BIB):end

=> d his]
'HIS]' IS NOT A VALID FORMAT FOR FILE 'SCISEARCH'

The following are valid formats:

The default display format is BIB.

ABS ---- AB

ALL ---- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC, AB, CC, ST, STP, RF, RE

BIB ---- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC

CBIB --- CBIB

IABS --- AB

IALL --- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC, AB, CC, ST, STP, RF, RE

IBIB --- AN, GA, TI, AU, CS, CYA, SO, DT, FS, LA, REC

IND ---- CC, ST, STP, RF

SAM ---- AN, GA, TI, REC, CC, ST, STP, RF

SCAN --- AN, GA, TI, CC, ST, STP, RF (random display without answer numbers)

TRI ---- AN, GA, TI, REC, CC, ST, STP, RF

HIT ---- All fields containing hit terms

KWIC --- All hit terms plus 20 words on either side

OCC ---- List of display fields containing hit terms

Hit terms will be highlighted in all displayable fields except PY.

To display a particular field or fields, enter the display field codes. For a list of display field codes, enter 'HELP DFIELDS' at an arrow prompt (=>). Examples of formats include: 'BIB'; 'AB'; 'SO,ST'. You may specify the format fields in any order, and the information will be displayed in the same order as the format specification.

The same formats (except for HIT, KWIC, OCC, and SCAN) may be used with the DISPLAY ACC command to display the record for a specified Accession Number.

ENTER DISPLAY FORMAT (BIB):end

=> f hid L12 2734 HID

=> d his

(FILE 'HOME' ENTERED AT 13:15:31 ON 09 JUL 2004)

```
FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS'
     ENTERED AT 13:16:11 ON 09 JUL 2004
           2912 S KANE M?/AU OR HAGEL A?/AU OR DOMBKOWSKI A?/AU
L1
              0 S L1 AND DIFFERENTIAL DISPLAY
L2
              O S L1 AND EXPRESS##### GENE
L3
             56 S L1 AND MRNA
1.4
              0 S L4 AND DIFFENTIAL?
1.5
1.6
              0 S L1 AND EXPRESS? MRNA
L7
          20308 S DIFFERENTIAL DISPLAY OR (DIFFERENTIALLY EXPRESSED (5A) MRNA)
L8
              0 S L1 AND L7
L9
            128 S L7 AND (OLIGO-DT (5A) (PRIMER OR SEQUENC?))
L10
              0 S L9 AND (REVERSE TRASCRIB?)
L11
              1 S L9 AND ADAPTOR
L12
           2734 F HID
=> 19 and (restriction enzyme or restriction endonuclease or endonuclease)
L9 IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter
"HELP COMMANDS" at an arrow prompt (=>).
=> s 19 and (restriction enzyme or restriction endonuclease or endonuclease)
T.13
             9 L9 AND (RESTRICTION ENZYME OR RESTRICTION ENDONUCLEASE OR ENDON
=> dup rem 19
PROCESSING COMPLETED FOR L9
             58 DUP REM L9 (70 DUPLICATES REMOVED)
=> dum rel 113
DUM IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter
"HELP COMMANDS" at an arrow prompt (=>).
=> dup renm 113
ENTER REMOVE, IDENTIFY, ONLY, OR (?):113
'L104' IS NOT VALID HERE
Enter "REMOVE" to identify and remove duplicate answers.
Enter "IDENTIFY" to identify duplicate answers in the answer set.
Enter "ONLY" to identify and create an answer set containing only
duplicate records.
ENTER REMOVE, IDENTIFY, ONLY, OR (?):end
=> dup rem 113
PROCESSING COMPLETED FOR L13
L15
              5 DUP REM L13 (4 DUPLICATES REMOVED)
=> d ibib abs 115 1-5
      ANSWER 1 OF 5 BIOTECHDS COPYRIGHT 2004 THOMSON DERWENT/ISI on STN
ACCESSION NUMBER: 2004-02692 BIOTECHDS
                  New annealing control primer capable of improving primer
TITLE:
                  annealing specificity in association with the alteration of
                  primer annealing temperature, useful for selectively
                  amplifying a target nucleic acid sequence;
                     DNA primer for mRNA differential expression detection
AUTHOR:
                  CHUN J Y
PATENT ASSIGNEE:
                  SEEGENE INC
PATENT INFO:
                  WO 2003093509 13 Nov 2003
APPLICATION INFO: WO 2002-KR816 1 May 2002
                  WO 2002-816 1 May 2002; WO 2002-816 1 May 2002
PRIORITY INFO:
DOCUMENT TYPE:
                  Patent
LANGUAGE:
                  English
OTHER SOURCE:
                  WPI: 2004-022665 [02]
```

2004-02692 BIOTECHDS DERWENT ABSTRACT:

NOVELTY - Annealing control primer capable of improving primer annealing specificity in association with the alteration of primer annealing temperature comprises a 3' and 5'-end portions separated by at least 2 deoxyinosine residues, universal bases or non-discriminatory base analogs, is new.

DETAILED DESCRIPTION - Annealing control primer capable of improving primer annealing specificity in association with the alteration of primer annealing temperature comprises a 3' and 5'-end portions separated by at least 2 deoxyinosine residues, universal bases or non-discriminatory base analogs, where the presence of deoxyinosine group positioned between the 3' and 5'-end portions plays as a switch in controlling primer annealing to a template nucleic acid in association with annealing temperature during PCR in order to interrupt the annealing of the 5'-end portion and limit primer annealing to the 3'-end portion at a first annealing temperature. The 5'-end portion comprises a universal primer sequence and serves as a universal priming site for subsequent amplification of reaction product generated from the annealing and extension of the 3'-end portion sequence to the template nucleic acid with the annealing of the 3'-end portion bothered or interrupted at a second annealing temperature. INDEPENDENT CLAIMS are also included for the following: (1) a kit comprising the annealing control primer; (2) selectively amplifying a target nucleic acid sequence from a nucleic acid molecule or mixture of nucleic acids using annealing control primers; (3) detecting DNA complementary to differentially expressed

mRNA in two or more nucleic acid samples using annealing control primers; (4) amplifying a target cDNA fragment comprising 5'-end region corresponding to the 5'-end of mRNA using annealing control primers; (5) amplifying a population of full-length double-stranded cDNAs complementary to mRNAs using annealing control primers; where the cDNAs comprise the complete 5'-end sequence information of the mRNAs; and (6) amplifying 5'-enriched double-stranded cDNA molecules complementary to mRNA molecules using annealing control primers.

BIOTECHNOLOGY - Preferred Primer: The annealing control primer has the general formula 5'-dNx-dIy-dNz-3. dNx = represents the 5'-end portion and contains a preselected arbitrary nucleotide sequence; dNz = represents the 3'-end portion; dIy = represents a deoxyinosine group having at least 2 deoxyinosine residues; dN = represents a deoxyribonucleotide; x, y or z = independently represents an integer; x = is the number of nucleotides in the 5'-end portion (which is an integer of 15-60); y = is the number of deoxyinosine residues separating the 3' and 5'-end portions (which is at least 3 or an integer of 2-15); and z = is the number of nucleotides in the 3'-end portion (which is an integer of 6-30). The deoxyribonucleotides comprise dAMP, dTMP, cDMP, dGMP, modified nucleotides or non-natural nucleotides. DNx includes a sequence that is recognized by a restriction endonuclease. It comprises at least one nucleotide with a hapten group. DNz is complementary to a target sequence in the template nucleic acid or to a consensus sequence found in a gene family. It is a degenerate sequence comprising combinations of nucleotides encoding a predetermined amino acid sequence. It comprises at least one ribonucleotide. It is a random nucleotide sequence. It is a deoxythymidine nucleotide sequence. It comprises at least 10 contiquous deoxythymidine nucleotides having 3'-NV, where V is deoxyadenosine, deoxycytidine or deoxyguanosine, and N is deoxythymidine, deoxyadenosine, deoxycytidine or deoxyguanosine The template nucleic acid of the annealing control primer is mRNA or cDNA derived from mRNA, or is single or double stranded DNA. The first annealing temperature of the annealing control primer should be lower than the second annealing temperature or is 37-65 or 50-72 degreesC. Preferred Method: Selectively amplifying a target nucleic acid sequence from a nucleic acid molecule or mixture of nucleic acids using annealing control primers comprises carrying out a two-stage PCR comprising: (1) amplifying the target nucleic acid sequence in a first-stage PCR comprising at least two cycles of primer annealing,

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primer extending and denaturing, by annealing a pair of annealing control
primers to the target nucleic acid sequence at a first annealing
temperature under conditions sufficient for template driven enzymatic DNA
synthesis to occur; extending the primers to obtain first amplification
product; denaturing the first amplification product to obtain denatured
amplification product; and (2) re-amplifying the denatured amplification
product at the second annealing temperature, which is high stringent
conditions, in a second-stage PCR comprising at least one cycle of
annealing, primer extending and denaturing, by annealing universal
primers corresponding to the 5'-end portion sequences of the annealing
control primers to the 5'-end sequences of the denatured amplification
product generated by the annealing control primers from step (1) and
extending the primers to generate second amplification product. The
target nucleic acid sequence is DNA or RNA. The first-stage PCR is
repeated at least twice. The second-stage PCR is repeated at least 10
times. The first annealing temperature is at least 40 degreesC, while the
second annealing temperature is at least 50 degreesC. Detecting DNA
complementary to differentially expressed
mRNA in two or more nucleic acid samples using annealing control
primers comprises: (1) providing a first sample of nucleic acids
representing a first population of mRNA transcripts and a second sample
of nucleic acids representing a second population of mRNA transcripts;
(2) separately contacting each of the first and second nucleic acid
samples with a first annealing control primer; (3) reverse transcribing
the differentially expressed mRNA to which
the first annealing control primer hybridizes to produce a first
population of DNA strands that are complementary to the
differentially expressed mRNA in the first
nucleic acid sample to which the first annealing control primer
hybridizes, and a second population of DNA strands that are complementary
to the differentially expressed mRNA in the
second nucleic acid sample to which the first annealing control primer
hybridizes; (4) purifying and quantifying each of the first and second
populations of complementary DNA strands; (5) contacting each of the
first and second populations of complementary DNA strands with a second
annealing control primer at a first annealing temperature, where the
second annealing control primer has a hybridizing sequence sufficiently
complementary to the first and second populations of DNA strands; (6)
extending the second annealing control primer using DNA polymerase to
produce a second DNA strand complementary to the first and second
populations of DNA strands; (7) amplifying each second DNA strand
obtained from step (6) at a second annealing temperature, by at least one
PCR cycle to obtain the first and second populations of amplification
products using 2 universal primers; and (8) comparing the presence or
level of individual amplification products in the first and second
populations of amplification products. The method further comprises
isolating the amplified cDNA product and cloning the isolated cDNA
product into a vector. The comparison comprises resolving each of the
first and second populations of amplification products by gel
electrophoresis through an ethidium bromide-stained agarose gel and
comparing the presence or level of bands of a particular size. The
nucleotide sequence of each of the first and second annealing control
primers comprises at least one nucleotide with a hapten group. The
universal primers comprise a sequence having 21-22 bp. The first nucleic
acid sample comprises mRNA expressed in a first cell and the second
nucleic acid sample comprises mRNA expressed in a second cell. The first
nucleic acid sample comprises mRNA expressed in a tumorigenic cell and
the second nucleic acid sample comprises mRNA expressed in a normal cell.
The first annealing control primer has the general formula of
5'-dN15-30-dI2-10-dT10-20-3'. dN = represents a deoxyribonucleotide and
contains preselected arbitrary nucleotide sequence; dI = represents a
deoxyinosine, universal base or non-discriminatory base analog; is
1-(2'-deoxy-beta-D-ribofuranosyl)-3-nitropyrole or 5-Nitroindole; dT =
represents a deoxythymidine; further comprises 3'-V. The first annealing
control primer comprises a sequence having comprises a sequence having 37
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bp. The second annealing control primer has the general formula of
5'-dN15-30-dI2-10-dN8-15-3', where the formula follows the same rule of
the formula 5'-dNx-dIy-dNz-3. It comprises a sequence having 32-40 bp.
Amplifying a target cDNA fragment comprising 5'-end region corresponding
to the 5'-end of mRNA using annealing control primers comprises: (1)
contacting the mRNA with a conventional Oligo dT
primer or random primer as a cDNA synthesis primer
under conditions sufficient for template driven enzymatic DNA synthesis
to occur; (2) reverse transcribing the mRNA to which cDNA synthesis
primer hybridizes to produce first-strand cDNA that is complementary to
the mRNA to which the cDNA synthesis primer hybridizes, resulting in
forming mRNA-cDNA intermediate; (3) permitting cytosine residues to be
tailed at the 3'-end of the first strand cDNA by the terminal transferase
reaction of reverse transcriptase in the presence of manganese under the
form of the mRNA-cDNA intermediate; (4) contacting a first annealing control primer to the cytosine tail at the 3'-end of the first cDNA
strand in the form of the mRNA-cDNA intermediate; where the first
annealing control primer comprises at least 3 guanine residues at its
3'-end to hybridize the cytosine tail of the 3'-end of the first cDNA
strand; (5) extending the tailed 3'-end of the first strand cDNA to
generate an additional sequence complementary to the first annealing
control primer using reverse transcriptase; where the first annealing
control primer is used as a template in the extension reaction; (6)
synthesizing the second-strand cDNA of the extended first-strand cDNA
using a universal primer by at least one cycle of PCR; where the
universal primer has a sequence complementary to the 5'-end extended
sequence of the first-strand cDNA; (7) synthesizing a target cDNA strand
using a second annealing control primer at a first annealing temperature
by at least one PCR cycle; and (8) amplifying the target cDNA strand
using 2 universal primers at a second annealing temperature, which is
high stringent conditions, by at least one PCR cycle; where the universal
primers have sequences complementary to both 3'- and 5'-ends of the
target cDNA strand, which comprises the sequences of the first and second
annealing control primers at both 3'- and 5'-ends. Amplifying a
population of full-length double-stranded cDNAs complementary to mRNAs
using annealing control primers; where the cDNAs comprise the complete
5'-end sequence information of the mRNAs comprises: (1) contacting the
mRNAs with a first annealing control primer under conditions sufficient
for template driven enzymatic DNA synthesis to occur, where the first
annealing control primer comprises a hybridizing sequence at 3'-end
portion complementary to the polyA tail of the mRNAs to hybridize; (2)
reverse transcribing the mRNAs to which the first annealing control
primer hybridizes to produce first strand cDNA sequences that are
complementary to the mRNAs to which the first annealing control primer
hybridizes, resulting in forming mRNA-cDNA intermediates; (3) permitting
cytosine residues to be tailed at the 3'-end of the first strand cDNAs by
the terminal transferase reaction of reverse transcriptase in the
presence of manganese under the form of the mRNA-cDNA intermediates; (4)
contacting a second annealing control primer to the cytosine tails at the
3'-end of the first cDNA strands in the form of the mRNA-cDNA
intermediates, where the second annealing control primer comprises at
least 3 guanine residues at its 3'-end to hybridize the cytosine tails of
the 3'-end of the first cDNA strands; (5) extending the tailed 3'-ends of
the first strand cDNAs to generate additional sequences complementary to
the second annealing control primer using reverse transcriptase, where
the second annealing control primer is used as a template in the
extension reaction; and (6) amplifying the extended first strand cDNAs
using 2 universal primers to obtain amplification products of full-length
cDNAs complementary to the mRNAs, by at least one cycle of PCR. The
method further comprises introducing the double-stranded cDNA molecules
obtained from step (6) into vectors. Amplifying 5'-enriched
double-stranded cDNA molecules complementary to mRNA molecules using
annealing control primers comprises: (1) contacting the mRNA molecules
with a first annealing control primer under conditions sufficient for
template driven enzymatic DNA synthesis to occur; where the first
```

annealing control primer comprises at least 6 random nucleotide sequences at 3'-end portion; and (2) performing steps (2)-(6) to amplify 5'-enriched double-stranded cDNA molecules.

USE - The annealing control primer is useful for selectively amplifying a target nucleic acid sequence from a nucleic acid molecule or mixture of nucleic acids, for detecting DNA complementary to differentially expressed mRNA in two or more nucleic acid samples or for amplifying a population of full-length double-stranded cDNAs complementary to mRNAs using annealing control primers.

ADVANTAGE - Primer annealing specificity is improved by the effect of the deoxyinosine residue group on the annealing of 3'- and 5'-end portions of ACP in accordance with the alteration of annealing temperature, which requires two stage PCR amplifications. Amplification of non-specific PCR products is interrupted by two-stage PCR amplifications, which are performed at low and high stringent conditions. Mispriming which is a major cause of false product amplification during PCR can be significantly minimized. The efficiency of PCR amplification is increased, which makes it easier to detect rare mRNAs. The reproducibility of PCR products is increased, which saves a great amount of time and cost. Agarose gel electrophoresis followed by ethidium bromide staining can be used for detecting differentially displayed RT-PCR products. The background problems arising from contamination of the primers used for cDNA synthesis for 5'- or 3'-rapid amplification of cDNA ends can be eliminated.

EXAMPLE - No relevant examples given. (136 pages)

L15 ANSWER 2 OF 5 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER:

1999:538415 CAPLUS

DOCUMENT NUMBER:

132:742

TITLE:

A Novel Strategy for Identifying Differential Gene

Expression: An Improved Method of Differential

Display Analysis

AUTHOR (S):

Kohroki, Junya; Tsuchiya, Mikako; Fujita, Sayaka; Nakanishi, Tsuyoshi; Itoh, Norio; Tanaka, Keiichi

CORPORATE SOURCE:

Department of Toxicology, Graduate School of Pharmaceutical Sciences, Osaka University, Suita,

Osaka, 565-0871, Japan

SOURCE:

Biochemical and Biophysical Research Communications

(1999), 262(2), 365-367

CODEN: BBRCA9; ISSN: 0006-291X

PUBLISHER:

Academic Press

DOCUMENT TYPE: LANGUAGE:

Journal English

We propose a novel alternative approach, an advanced method for recently developed strategies, for identifying differentially expressed genes. Firstly, double-stranded cDNAs were digested using Sau3AI and the 3'-end restriction fragments of the cDNA were ligated to a double-stranded adapter. Next, the restriction fragments were directly amplified using several combinations of adapter-specific primers and FITC-labeled oligo dT primers. The selected cDNA fragments were displayed on a polyacrylamide Neither nested PCR nor purification of 3'-end fragments are necessary. We examined the validity of this approach by evaluating gene expression changes during granulocytic differentiation of HL-60 cells. This method can theor. detect almost all gene expression changes more rapidly and through simpler manipulations than by any other approach. (c) 1999 Academic Press.

REFERENCE COUNT:

11 THERE ARE 11 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L15 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2004 ACS on STN

ACCESSION NUMBER:

1999:704522 CAPLUS

DOCUMENT NUMBER:

132:132862

TITLE:

A Novel Strategy for Identifying Differential Gene Expression: An Improved Method of Differential

Display Analysis. [Erratum to document cited

in CA132:7421

AUTHOR (S): Kohroki, Junya; Tsuchiya, Mikako; Fujita, Sayaka;

Nakanishi, Tsuyoshi; Itoh, Norio; Tanaka, Keiichi

CORPORATE SOURCE: Dep. Toxicology, Graduate School Pharmaceutical

> Sciences, Osaka Univ., Suita, Osaka, 565-0871, Japan Biochemical and Biophysical Research Communications

(1999), 265(1), 272

CODEN: BBRCA9; ISSN: 0006-291X

Academic Press PUBLISHER:

DOCUMENT TYPE: Journal LANGUAGE: English

SOURCE:

On page 365, column 2, line 18, and on page 366, column 2, line 13 to page 367, column 1, line 1, the sequence of the adapter primer was incorrect as printed. The sequence should read "(5'-GTCGACATGAGTGTGATCN1N2-3',..." instead of "(5'-GTCGACATGAGTGTGAN1N2-3',..." and "5'-GTCGACATGAGTGTGANN-3'...", resp., as printed. (c) 1999 Academic Press.

MEDLINE on STN L15 ANSWER 4 OF 5 DUPLICATE 1

ACCESSION NUMBER: 2000064167 MEDLINE DOCUMENT NUMBER: PubMed ID: 10596370

TITLE: The coupling of differential display

and AFLP approaches for nonradioactive mRNA fingerprinting.

AUTHOR: Ivashuta S; Imai R; Uchiyama K; Gau M

Hokkaido National Agricultural Experiment Station, Sapporo, CORPORATE SOURCE:

Japan.. sergey@cryo.affrc.go.jp

SOURCE: Molecular biotechnology, (1999 Sep) 12 (2) 137-41.

Journal code: 9423533. ISSN: 1073-6085.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199912

ENTRY DATE: Entered STN: 20000114

> Last Updated on STN: 20000114 Entered Medline: 19991230

AB We have modified the differential display of 3'-end restriction fragments of cDNA technique by combining it with the amplified

fragment length polymorphism (AFLP) approach and silver staining. Modified oliqo-dT primers were used for a reverse transcription step. ds

cDNA was digested with the Mse I restriction enzyme

and then ligated with an AFLP adapter. The modified template was amplified with oligo-dT primers in a preamplification step (asymmetric PCR) that enriched the template for 3'-end sequences; subsequently, the enriched template was amplified with an AFLP primer having a selective extension and an anchored oligo-dT primer

(conventional PCR step). We demonstrated that the asymmetric preamplification step facilitates the preferential amplification of 3'-end fragments and the resulting PCR products can be clear resolved on silver-stained gel. The presented procedure takes advantages of silver-stained gels, generates reproducible display patterns, and allows reliable reamplification of isolated fragments which contain both upstream and downstream primer sequences.

L15 ANSWER 5 OF 5 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN

ACCESSION NUMBER: 95:703492 SCISEARCH

THE GENUINE ARTICLE: RY760

DESCRIPTION OF THE ENTIRE MESSENGER-RNA POPULATION BY A TITLE

3'-END CDNA FRAGMENT GENERATED BY CLASS IIS RESTRICTION

ENZYMES

AUTHOR: KATO K (Reprint)

JRDC, ERATO, OKAYAMA CELL SWITCHING PROJECT, SAKYO KU, CORPORATE SOURCE:

103-5 TANAKAMONZENCHO, KYOTO 606, JAPAN (Reprint)

COUNTRY OF AUTHOR: JAPAN

SOURCE: NUCLEIC ACIDS RESEARCH, (25 SEP 1995) Vol. 23, No. 18, pp. 3685-3690.

ISSN: 0305-1048. Article; Journal

DOCUMENT TYPE: FILE SEGMENT:

LIFE

LANGUAGE:

ENGLISH

REFERENCE COUNT:

17

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

AB A novel means of recording the expression status of the total g

A novel means of recording the expression status of the total gene population is described, Digestion of cDNA by class IIS restriction enzymes produces a fragment with a poly (A) stretch and a 5' overhang with an unknown sequence, This fragment contains information such as the class IIS enzyme that cuts cDNA nearest to the poly (A) stretch, the sequence of the 5' overhang, and the size of the fragment. Expressed genes can be discriminated and displayed by the fragment as follows: (i) cut the cDNA with one class IIS restriction enzyme; (ii) ligate the digested cDNA to that from a pool of 64 biotinylated adaptors cohesive to all possible overhangs; (iii) digest by other two class IIS enzymes; (iv) recover the ligated molecules with streptavidin-coated paramagnetic beads; (v) perform PCR with the adaptor-primer and an anchored oligo-dT primer; (vi) separate the amplified fragments by denaturing polyacrylamide gel electrophoresis. Repeat the experiment with 64 adaptors, three enzymes and three anchored oligo-dT primers displays most of the expressed genes, Because redundancy is minimized, this technique is also ideal for generating tags for expressed genes, with which to construct a transcript map of the genome.

#### => d his

L1

L3

L5

(FILE 'HOME' ENTERED AT 13:15:31 ON 09 JUL 2004)

FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS' ENTERED AT 13:16:11 ON 09 JUL 2004

2912 S KANE M?/AU OR HAGEL A?/AU OR DOMBKOWSKI A?/AU

L2 0 S L1 AND DIFFERENTIAL DISPLAY

0 S L1 AND EXPRESS##### GENE

L4 56 S L1 AND MRNA

0 S L4 AND DIFFENTIAL?

L6 0 S L1 AND EXPRESS? MRNA

L7 20308 S DIFFERENTIAL DISPLAY OR (DIFFERENTIALLY EXPRESSED (5A) MRNA)

L8 0 S L1 AND L7

L9 128 S L7 AND (OLIGO-DT (5A) (PRIMER OR SEQUENC?))

L10 0 S L9 AND (REVERSE TRASCRIB?)

L11 1 S L9 AND ADAPTOR

L12 2734 F HID

L13 9 S L9 AND (RESTRICTION ENZYME OR RESTRICTION ENDONUCLEASE OR EN

L14 58 DUP REM L9 (70 DUPLICATES REMOVED)

L15 5 DUP REM L13 (4 DUPLICATES REMOVED)

=> s 19 and (linear amplification or selective amplification)

L16 5 L9 AND (LINEAR AMPLIFICATION OR SELECTIVE AMPLIFICATION)

#### => d ibib abs 116 1-5

L16 ANSWER 1 OF 5 MEDLINE ON STN ACCESSION NUMBER: 96216735 MEDLINE DOCUMENT NUMBER: PubMed ID: 8645264

TITLE: Combining the technique of RNA fingerprinting and

differential display to obtain differentially expressed mRNA.

AUTHOR: Diachenko L B; Ledesma J; Chenchik A A; Siebert P D CORPORATE SOURCE: CLONTECH Laboratories, Inc., Palo Alto, California

94303-4230, USA.

SOURCE: Biochemical and biophysical research communications, (1996

Feb 27) 219 (3) 824-8.

Journal code: 0372516. ISSN: 0006-291X.

PUB. COUNTRY: United States

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals

ENTRY MONTH: 199607

ENTRY DATE: Entered STN: 19960726

Last Updated on STN: 19960726 Entered Medline: 19960715

AB We have modified recently developed methods of RNA fingerprinting and differential display based on arbitrarily primed PCR which can be used for detection and cloning of differentially expressed mRNAs. Our protocol requires only a single cDNA synthesis for each different RNA sample, in contrast to the multiple cDNA reactions required for differential display method, followed by

selective amplification of cDNA sequence

fraction by arbitrary and <code>oligo(dT)</code> primers. We have shown that the longer primers (25-29-mers) allow the use of optimal dNTP concentration and higher stringency PCR. Further improvements include using TaqStart antibody for hot start PCR and thermostable enzyme mixes suitable for long-distance PCR. Long-distance PCR enables the method to display bands of up to 2 kb and should result in a higher fidelity of PCR products to the original RNA template. When these improvements are combined the resulting method is highly reproducible, and more than 85% of the differentially expressed bands can be confirmed by Northern blot analysis.

L16 ANSWER 2 OF 5 EMBASE COPYRIGHT 2004 ELSEVIER INC. ALL RIGHTS RESERVED.

on STN

ACCESSION NUMBER: 96111972 EMBASE

DOCUMENT NUMBER: 1996111972

TITLE: Combining the technique of RNA fingerprinting and

differential display to obtain differentially expressed mRNA.

AUTHOR: Diachenko L.B.; Ledesma J.; Chenchik A.A.; Siebert P.D.

CORPORATE SOURCE: CLONTECH Laboratories Inc, 1020 East Meadow Circle, Palo

Alto, CA 94303-4230, United States

SOURCE: Biochemical and Biophysical Research Communications, (1996)

219/3 (824-828).

ISSN: 0006-291X CODEN: BBRCA

COUNTRY: United States
DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 022 Human Genetics

029 Clinical Biochemistry

LANGUAGE: English SUMMARY LANGUAGE: English

AB We have modified recently developed methods of RNA fingerprinting and differential display based on arbitrarily primed PCR which can be used for detection and cloning of differentially expressed

mRNAs. Our protocol requires only a single cDNA synthesis for each different RNA sample, in contrast to the multiple cDNA reactions required

for differential display method, followed by

selective amplification of cDNA sequence
fraction by arbitrary and oligo(dT) primers. We have

shown that the longer primers (25-29-mers) allow the use of optimal dNTP concentration and higher stringency PCR. Further improvements include using TaqStart antibody for hot start PCR and thermostable enzyme mixes suitable for long-distance PCR. Long-distance PCR enables the method to display bands of up to 2 kb and should result in a higher fidelity of PCR products to the original RNA template. When these improvements are combined the resulting method is highly reproducible, and more than 85% of the differentially expressed bands can be confirmed by Northern blot analysis.

ACCESSION NUMBER: 1996:219632 BIOSIS DOCUMENT NUMBER: PREV199698775761

TITLE: Combining the technique of RNA fingerprinting and

differential display to obtain differentially expressed mRNA.

AUTHOR(S): Diachenko, Luda B.; Ledesma, John; Chenchik, Alex A.;

Siebert, Paul D.

CORPORATE SOURCE: CLONTECH Lab. Inc., 1020 East Meadow Circle, Palo Alto, CA

94303-4230, USA

SOURCE: Biochemical and Biophysical Research Communications, (1996)

Vol. 219, No. 3, pp. 824-828. CODEN: BBRCA9. ISSN: 0006-291X.

DOCUMENT TYPE: Article LANGUAGE: English

ENTRY DATE: Entered STN: 8 May 1996

Last Updated on STN: 8 May 1996

AB We have modified recently developed methods of RNA fingerprinting and differential display based on arbitrarily primed PCR which can be used for detection and cloning of differentially expressed

mRNAs. Our protocol requires only a single cDNA synthesis for each different RNA sample, in contrast to the multiple cDNA reactions required for **differential display** method, followed by

selective amplification of cDNA sequence

fraction by arbitrary and oligo (dT) primers. We have shown that the longer primers (25-29-mers) allow the a

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L16 ANSWER 4 OF 5 SCISEARCH COPYRIGHT 2004 THOMSON ISI on STN

ACCESSION NUMBER: 96:211116 SCISEARCH

THE GENUINE ARTICLE: TZ799

TITLE: COMBINING THE TECHNIQUE OF RNA FINGERPRINTING AND

DIFFERENTIAL DISPLAY TO OBTAIN

DIFFERENTIALLY EXPRESSED MESSENGER-RNA

AUTHOR: DIACHENKO L B (Reprint); LEDESMA J; CHENCHIK A A; SIEBERT

Рυ

CORPORATE SOURCE: CLONTECH LABS INC, 1020 E MEADOW CIRCLE, PALO ALTO, CA,

94303 (Reprint)

COUNTRY OF AUTHOR: USA

SOURCE: BIOCHEMICAL AND BIOPHYSICAL

BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, (27

FEB 1996) Vol. 219, No. 3, pp. 824-828.

ISSN: 0006-291X.

DOCUMENT TYPE:

Article; Journal LIFE

FILE SEGMENT: LANGUAGE:

ENGLISH

REFERENCE COUNT:

12

REFERENCE COUNT: 12

\*ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS\*

B We have modified recently developed methods of RNA fingerprinting and differential display based on arbitrarily primed PCR which can be used for detection and cloning of differentially expressed mRNAs. Our protocol requires only a single cDNA synthesis for each different RNA sample, in contrast to the multiple cDNA reactions required for differential display method, followed by

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fraction by arbitrary and <code>oligo(dT)</code> primers. We have shown that the longer primers (25-29-mers) allow the use of optimal dNTP concentration and higher stringency PCR. Further improvements include using TaqSart antibody for hot start PCR and thermostable enzyme mixes suitable for long-distance PCR. Long-distance PCR enables the method to

display bands of up to 2 kb and should result in a higher fidelity of PCR products to the original RNA template. When these improvements are combined the resulting method is highly reproducible, and more than 85% of the differentially expressed bands can be confirmed by Northern blot analysis. (C) 1996 Academic Press, Inc.

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L16 ANSWER 5 OF 5 CAPLUS COPYRIGHT 2004 ACS on STN ACCESSION NUMBER: 1996:162465 CAPLUS DOCUMENT NUMBER: 125:106264
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TITLE: Combining the technique of RNA fingerprinting and

differential display to obtain

differentially expressed

mRNA

AUTHOR(S): Diachenko, Luda B.; Ledesma, John; Chenchik, Alex A.;

Siebert, Paul D.

CORPORATE SOURCE: CLONTECH Laboratories, Inc., Palo Alto, CA,

94303-4230, USA

SOURCE: Biochemical and Biophysical Research Communications

(1996), 219(3), 824-28

CODEN: BBRCA9; ISSN: 0006-291X

PUBLISHER: Academic DOCUMENT TYPE: Journal LANGUAGE: English

AB We have modified recently developed methods of RNA fingerprinting and differential display based on arbitrarily primed PCR

which can be used for detection and cloning of differentially expressed mRNAs. Our protocol requires only a single cDNA synthesis for each different RNA sample, in contrast to the multiple cDNA reactions required

for differential display method, followed by

selective amplification of cDNA sequence

fraction by arbitrary and oligo(dT) primers. We have shown that the longer primers (25-29-mers) allow the use of optimal dNTP concentration and higher stringency PCR. Further improvements include using

Concentration and higher stringency PCR. Further improvements include using TaqStart antibody for hot start PCR and thermostable enzyme mixes suitable for long-distance PCR. Long-distance PCR enables the method to display bands of up to 2 kb and should result in a higher fidelity of PCR products to the original RNA template. When these improvements are combined the resulting method is highly reproducible, and more than 85% of the differentially expressed bands can be confirmed by Northern blot anal.

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L13

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FILE 'MEDLINE, BIOTECHDS, EMBASE, BIOSIS, SCISEARCH, CANCERLIT, CAPLUS' ENTERED AT 13:16:11 ON 09 JUL 2004

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L1 2912 S KANE M?/AU OR HAGEL A?/AU OR DOMBKOWSKI A?/AU L2 0 S L1 AND DIFFERENTIAL DISPLAY
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0 S L1 AND EXPRESS##### GENE

L4 56 S L1 AND MRNA

0 S L4 AND DIFFENTIAL?

L6 0 S L1 AND EXPRESS? MRNA

L7 20308 S DIFFERENTIAL DISPLAY OR (DIFFERENTIALLY EXPRESSED (5A) MRNA)

L8 0 S L1 AND L7

L9 128 S L7 AND (OLIGO-DT (5A) (PRIMER OR SEQUENC?))

L10 0 S L9 AND (REVERSE TRASCRIB?)

L11 1 S L9 AND ADAPTOR

L12 2734 F HID

9 S L9 AND (RESTRICTION ENZYME OR RESTRICTION ENDONUCLEASE OR EN

L14 58 DUP REM L9 (70 DUPLICATES REMOVED)

L15 5 DUP REM L13 (4 DUPLICATES REMOVED)

L16 5 S L9 AND (LINEAR AMPLIFICATION OR SELECTIVE AMPLIFICATION)

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=> s 117 and differential display
           271 L17 AND DIFFERENTIAL DISPLAY
=> s l18 and oligo dT
            27 L18 AND OLIGO DT
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               ENDONUCLEASE)
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     Differential display using one-base anchored
     oligo-dT primers.
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     Liang P.; Zhu W.; Zhang X.; Guo Z.; O'Connell R.P.; Averboukh
     L.; Wang F.; Pardee A.B.
CS
     Division Cell Growth and Regulation, Dana-Farber Cancer Institute, Boston,
     MA 02115, United States
SO
     Nucleic Acids Research, (1994) 22/25 (5763-5764).
     ISSN: 0305-1048 CODEN: NARHAD
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     029
             Clinical Biochemistry
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     article
     cell transformation
     controlled study
     dna sequence
     fibroblast
     gene amplification
     molecular cloning
     nonhuman
     northern blotting
     polymerase chain reaction
     priority journal
     rat
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     Drug Descriptors:
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     polyadenylic acid
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       restriction endonuclease
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